REMARKS/ARGUMENTS

1. Claims 1-6 and 9-10 are pending in the Application. Applicant has cancelled Claims 1-6 and 9-10, without waiver or prejudice, and has presented new claims 11-20.

2. Specification. Changes made to the Specification on pages 11 and 23 correct clear inadvertent, typographical and editorial errors. Specifically, page 12, the addition of "μM" after the number 3 at the end of each of lines 8 and 9 corrects the inadvertent omission of the concentration unit. As the Examiner will appreciate, this correction is fully supported by the Specification as originally filed; hence, no new matter is being presented by this amendment. A concentration unit must be present after the number 3 in each of lines 8 and 9 because X is a concentration (see, for example, page 12, line 1 of the Specification). Second, 3 μM is taught to be a preferred concentration (see, for example, page 21, lines 10-11 of the Specification).

3. Enablement. Examiner rejected claims 1-6 and 9-10 under 35 USC 112, first paragraph, as failing to comply with the enablement requirement. Examiner states, "neither the specification nor the prior art teach how to use an equation to predict the IC_{50} of a compound at a single concentration wherein a percent response (e.g., inhibition) is not measured at that concentration." Applicants certainly agree that the prior art does not teach this. That is why all the claims are patentable over the art. However, Applicants vigorously disagree with the Examiner as to the specification not teaching how to use such an equation to predict IC_{50}.

The present invention provides methods to accurately and rapidly predict the IC_{50} of unknown compounds from just a single data point for each compound. As disclosed in the Specification, the IC_{50} of a compound is its concentration at which 50% of an effect (response) is inhibited (Specification, page 8 lines 5-6). The effect may be the production of an analyte in a biological assay.

Specifically, in the Specification, the biological assay is performed using the target on one or more test (unknown) compounds at concentration X to determine the percent inhibition for each test compound at concentration X and the predetermined mathematical relationship is then used to calculate the IC_{50} for each test compound from the percent inhibition determined at concentration X for that compound. The calculation is quite simple. The percent inhibition is "plugged into" the simple mathematical relationship

$$IC_{50} = \exp\{a + b \cdot (\text{percent inhibition at concentration X})\}$$

where a and b are constants that were determined when the mathematical relationship was determined.

As explained in the Specification, in vivo drug interaction with the body's cytochrome P450 (CYP) is clinically important, and in vitro methods for predicting such interaction have become more important as the number of compounds to be screened for potential use as drugs has increased (Specification, page 1, line 21, to page 2, line 2). The IC_{50} of a compound in the appropriate biological assay against a given target (e.g., CYP) may be indicative of potential in vivo drug interaction.

Compounds are typically tested to determine their IC_{50} using at least (and usually at more than) two different concentrations, and a curve is drawn by fitting the concentration-effect data points to a mathematical model. Once this dose response curve for a compound of interest is known (e.g., the equation of the curve has been determined), the concentration corresponding to a 50% reduction in the maximum effect (i.e., IC_{50}) is easily determined from the curve. Moody et al., "Fully Automated Analysis Of Activities Catalysed By The Major Human Liver Cytochrome P450 (CYP) Enzymes: Assessment Of Human CYP Inhibition Potential," Xenobiotica, volume 29, no. 1, pages 53-75 (1999), shows use of seven points in some instances and two points in other instances for determining the dose response curve for a compound.

Applicants' methods, which requires only a single data point for a compound to predict its IC_{50}, represents a significant advance in the art. The single data point used in Applicants' method is the effect (inhibition) observed at a preselected concentration X. Thus, with a biological screening assay, each compound of interest needs to be put into only one reaction mixture (e.g., in a single microwell), which mixture includes the other materials (e.g., compounds and cells) used in the assay, to allow the assay to be run and the level of effect (response) to be measured. When a reaction mixture does not contain any inhibitory compound, the maximum effect (response) is achieved and can be measured. The percent inhibition for any compound at a given concentration X may be obtained by dividing the maximum effect into the effect observed for the compound at the given concentration X and subtracting the result from 100%.

Applicants' methods for determining the IC_{50} of an unknown compound using just a single data point (i.e., the measured effect for the unknown compound at concentration X) makes use of a mathematical relationship between IC_{50} and measured effect at the preselected concentration X for a group of known compounds. The relationship must be determined before it can be used to predict IC_{50} for an unknown compound from its measured response (effect) in the assay at concentration X. The relationship need not be determined immediately before each unknown compound is assayed. In fact, one of the features of Applicants' invention is that once

the relationship between IC_{50} and measured effect at preselected (standardized) concentration X has been determined, it need not be determined again.

Broadly speaking, the steps for determining the mathematical relationship involve selecting a biological assay and then running the assay on known compounds, obtaining (measuring) the effect (response) at each of several concentrations (and by simple division and subtraction, converting each measured effect to percent inhibition), and determining the curve relating concentration to percent inhibition (i.e., the dose response curve) for that compound. The curve for each compound is determined by fitting the data for that compound to a model, for example, the Hill function of formula I:

$$\text{percent inhibition} = \frac{100}{1 + \left(\frac{IC_{50}}{\text{concentration}}\right)^{h}}$$

where h is a constant (e.g., -1). As is known by those skilled in the art, the data may be fit to other models (see, e.g., the above-cited Moody et al. article, which at page 57 indicates that those skilled in the art know such data can be fit to more than one formula).

Once the dose response curve has been obtained for each known compound, two different points on the curve are identified: (1) the IC_{50} point (the point at which the response (effect) is 50% of the maximum response (again, the maximum response is obtained when no inhibitory compound is present)) and (2) the percent inhibition at preselected concentration X. Concentration X may be any value, consistent with the assay, the amount of each compound available, etc., but once it has been selected, it becomes a fixed value. For screening drug candidates, which typically are produced in small quantities, concentrations in the micromolar (μM) range, e.g., 1 to 10 μM, are usually satisfactory. Example 3 of the Application discusses the method Applicants use to select 3 μM from among the three concentrations X being considered for the assay of interest (i.e., 1 μm, 3 μM, and 10 μM). The set of pairs of values for each known compound (each pair being the compound's IC_{50} concentration and its percent inhibition at concentration X) is then fit to a curve of formula II:

$$IC_{50} = \exp\{a + b \cdot (\text{percent inhibition at concentration X})\}$$

where a and b are constants determined during the curve fitting process. Examples 4, 5, and 6 illustrate determination of specific formula II (i.e., where constants a and b have been

determined). Example 6 (see Figure 11) shows that more than one formula II may be used to embody a single set of data.

Once this relationship has been determined, Applicants' method of predicting IC_{50} for unknown compounds can be performed. Thus, in Applicants' method, unknown compounds are tested at the preselected and previously used concentration X against the same target in the same assay as was used to develop the relationship of formula II, the response (effect) is measured and converted by simple arithmetic to its equivalent, percent inhibition, and formula II is used to calculate a predicted IC_{50} for that compound. The examples of the Application demonstrate the high accuracy of the method.

Moreover, Applicants disagree with the Examiner's assertion on page 4 of the Official Action that "[t]he specification does not disclose any working examples for actually determining an IC_{50} of a 'remaining or future (unknown?) compound at a single concentration using an equation as determined by claimed step (d)." Applicants also disagree with the Examiner's assertion that "[t]he specification does not specifically provide guidance for predicting an IC_{50} of a compound using an equation obtained through the claimed method steps" (id.).

The application unquestionably does contain examples and provide sufficient guidance. See, e.g., Specification, page 14, lines 8-9 and lines 11-14; page 15, lines 10-11; page 18, lines 29-31; page 21, lines 1-3, lines 5-6, and lines 10-11; page 22, lines 17-18 and lines 27-28; page 23, line 5; and page 20, lines 4-7 (emphasis added):

> To perform the test we randomly selected 103 data points from the 163 available data points as a training set, while using the remaining 60 data points as a test set. Models were then built using the training set and were followed by predicting the $\log_{10}(IC_{50})$ of the test set.

Predicting the common logarithm of IC_{50} is the same as predicting IC_{50}.

Accordingly, entry of the amendments hereinabove and reconsideration of the Office Action mailed June 17, 2003 are respectfully requested.

4. Written Description. Claims 9-10 are rejected under 35 USC 112, first paragraph, as failing to comply with the written description requirement. Examiner states that "[t]he claim(s) contains subject matter which was not described in the specification in such a way as to reasonably convey to one skilled in the relevant art that the inventor(s), at the time the application was filed had possession of the claimed invention."

Applicant believes that it has fully complied with the written description requirement. Specifically, under written description, Applicant believes that newly submitted claims 11-20 fully supported and described in the Specification in such a way as to reasonably convey to one skilled in the relevant are that the inventors, at the time of the Application was filed, had possession of the claimed invention. Therefore, Applicants request that the rejection for inadequate written description be withdrawn.

Cancelled claim 1 has been rewritten as new claim 11. As set forth in that new claim, after the mathematical relationship between IC_{50} and percent inhibition at concentration X has been determined (steps (a) through (d)), that relationship is used to predict the IC_{50} value for an unknown compound using just one piece of information, namely, the percent inhibition at concentration X determined using the same target in the same assay as was used to derive the mathematical relationship.

The first four paragraphs ((a) through (d)) are based, respectively, upon paragraphs (a) to (d) of canceled claim 1, with minor changes for further clarity and consistency. Thus, for example, "developing" has been added to "identifying" at the start of paragraph (a). That change finds support in the application as filed (Specification, e.g., page 11, line 14).

"EC_{50}" has been removed from the rewritten claim because "IC_{50}" is defined in the application to include "EC_{50}" (Specification, e.g., page 11, lines 6-7). "Percent effect" has also been removed from the claim for further clarity so that the claim explicitly mentions only "percent inhibition" and not both "percent effect" and "percent inhibition." As is known to those skilled in the art, and as is abundantly clear from the application, the two are complementary, together totaling 100% (of the maximum effect). "Using a computer" has been deleted from the claim. Those skilled in the art can (and know how to) choose or not choose to use a computer, depending on how much data is being manipulated, the speed at which an answer is desired, etc. There is nothing in the invention per se that requires the use of a computer in all cases and the claims need not be so limited.

The substance of canceled claim 3 has been incorporated into paragraph (d) of claim 11. In canceled claim 1, the first portion of paragraph (e) was redundant in view of paragraph (d). The redundancy has been eliminated in new claim 11. Finally, the last portion of paragraph (e) of canceled claim 1 (i.e., determining IC_{50}) has been expanded and clarified and now appears in paragraphs (e) and (f) of new claim 11.

These changes are believed to obviate all of the Examiner's Section 112, second paragraph, rejections detailed in the Official Action at pages 5-7.

Claim 12 is essentially canceled claim 2 with minor changes for further clarity.

Claim 13 is a new independent claim similar to canceled claim 1 and to new claim 11. Claim 13 is directed to a method of predicting IC_{50} concentrations for test (unknown) compounds using a predetermined relationship between IC_{50} and percent inhibition at preselected concentration X where the relationship has been previously determined in the manner described above.

Paragraphs (i) through (iv) of claim 13 (which concern determining the relationship) are based, respectively, upon paragraphs (a) to (d) of canceled claim 1, with minor changes for further clarity and consistency, and are essentially the same as paragraphs (a) through (d) of new claim 11. Thus, as specified in new claim 13, after the mathematical relationship between IC_{50} and percent inhibition at concentration X has been determined by someone at some prior time, it is used to predict the IC_{50} value for an unknown compound using just one piece of information, namely, the percent inhibition at concentration X determined using the same target in the same assay as was used to derive the mathematical relationship. This is what is embodied in the method of claim 13 (steps (a) and (b)).

Temporally separating how the mathematical relationship was predetermined (items (i) through (iv)) from the method steps of first running the assay and measuring just a single data point for an unknown compound (the effect at concentration X) and then using the predetermined relationship to predict the unknown compound's IC_{50} (method steps (a) and (b)) is fully supported by the application as filed (see, e.g., Application, page 20, lines 6-7), is clearly patentable over the art, and gives applicants the protection to which they are entitled by preventing an infringer from stealing the heart of the invention while arguing that it does not infringe because it did not itself determine the mathematical relationship prior to using that relationship to predict an IC_{50} value from a single data point.

The method of claim 13 (steps (a) and (b)) for predicting IC_{50} is patentable because the art uses more than just a single data point (see, e.g., Moody et al.), the art does not teach the need to obtain the single data point at a preselected concentration X, and the art does not teach how a single response value from an assay for a compound could by itself be used to predict IC_{50}. Moreover, the method of claim 13 (steps (a) and (b)) for predicting IC_{50} is patentable because it requires use of the mathematical relationship that someone at some time in the past has

determined according to paragraphs (i) through (iv) of claim 13, a relationship that is not to applicants' knowledge disclosed or suggested in the art.

Claim 14 is essentially canceled claim 2 with minor changes for further clarity and is essentially the same as new claim 12.

Claim 15 is the third independent claim and is directed to the method of determining the mathematical relationship that can later be used to predict an IC_{50} value from a single data point. Steps (a) through (d) of claim 15 are based, respectively, upon paragraphs (a) through (d) of canceled claim 1, with minor changes for further clarity and consistency, and are essentially the same as paragraphs (a) through (d) of new claim 11. Claim 15 is patentable because, as far as is known by applicants, the art does not teach or suggest the mathematical relationship or the steps required to obtain that relationship or that the relationship can later be used to predict IC_{50} from just a single data point (i.e., the percent inhibition at the preselected concentration X, which concentration was used to obtain the mathematical relationship).

Claim 16 is essentially canceled claim 2 with minor changes for further clarity and is also essentially the same as new claims 12 and 14.

Claim 17 is essentially the same as canceled claim 4.

Claim 18 is essentially the same as canceled claim 5.

Claim 19 recites that the target is a biologically active protein and is fully supported by the application as filed (see, e.g., Application, page 11, lines 5-6).

Claim 20 depends from claim 19 and recites that the target is selected from the group consisting of enzymes, receptors, and transporters. The claim is fully supported by the application as filed (see, e.g., Application, page 11, lines 9-10) and is a combination of canceled claims 6, 7, and 8. Applicants specifically note that claims 7 and 8 were held withdrawn as being to non-elected species (Official Action, page 2).

Finally, applicants specifically note that all of the new claims are readable upon the elected species and that there are linking claims in application (e.g., the three independent claims).

5. Indefiniteness. With respect to Examiner's rejection of claims 1-6 and 9-10 under 35 USC 112, second paragraph, as being indefinite for failing to particularly point out and distinctly claim the subject matter which Applicant regards as the invention, Applicant believes that this rejection is moot in light of the fact that it has cancelled all pending claims in the Application, and has presented new claims 11-20 which clearly particularly point out and distinctly claim the subject matter of Applicant's invention, thus obviating this rejection.

6. Applicants believe that the cancellation of the presently pending claims 1-6 and 9-10, and the newly submitted claims 11-20 place the Application in condition for immediate allowance. Therefore, entry of the amendments hereinabove and reconsideration of the Office Action mailed June 17, 2003 are respectfully request. Such prompt and favorable action is earnestly solicited.

Respectfully submitted,

Date: Oct. 17, 2003

Pfizer Inc.
Patent Department, MS 8260-1611
Eastern Point Road
Groton, Connecticut 06340
(860) 715-5756

Irene M. Reininger
Attorney for Applicant(s)
Reg. No. 48,439